# Journal of Applied Microscopy

and

## Laboratory Methods

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# Journal of Applied Microscopy Laboratory Methods.

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# The New Biological Laboratories of Ripon College.



INGRAM HALL FROM THE NORTH.

The Biological Laboratories of Ripon College are located in the first story and basement of Ingram Hall, which was occupied for the first time at the beginning of the present school year.

Ingram Hall contains, besides the department of biology, the departments of physics and chemistry, physics being on the second floor and chemistry on the third floor. The building, which was named from the principal donor, Mr. O. H. Ingram of Eau Claire, is 73 by 121 feet in its outside dimensions, and is located at the brow of the hill on the campus in such a way that the south side

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is four stories in height. The longer dimension of the building is east and west —the entrance to the first floor being on the north side, and to the basement on the south side. The material of the building is dark red vitrified brick, with rustications of Roman brick and trimmings of Bedford stone. The building is in form a rectangle, with only such projections as are necessary to relieve the monotony of its exterior surface. The architect was instructed to make as many windows as the character of the structure would permit, and the result is that all the rooms are amply lighted. A feature of the construction is the character of the partitions. There are two solid brick partitions running through the whole height of the building. The other partitions are only two inches in thickness, the necessary support being given by heavy pillars. These partitions, a device of the architect, Mr. H. K. Holsman of Chicago, have a core of wood, which is plastered solid on both sides. At first thought, they would seem to be very unsubstantial, but as a matter of fact, after the adamant plaster is applied, not only are the walls firm and substantial, but sound is not carried through them to any disturbing extent. The manifest advantages of the partitions are the economy of floor space and the fact that they are practically fire-proof.

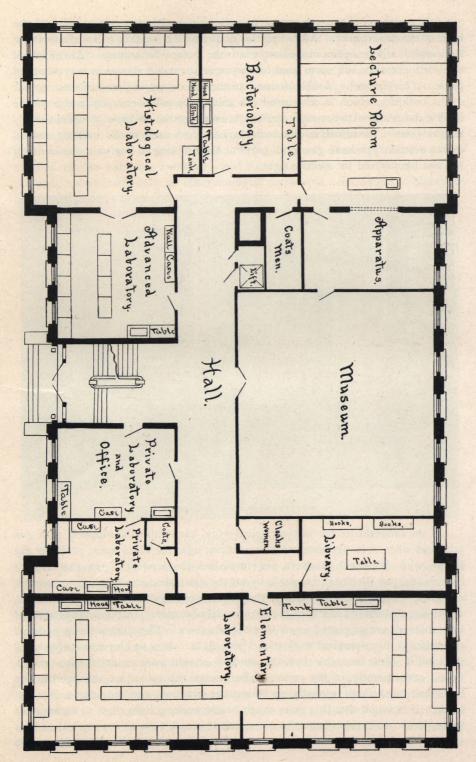
The building is finished in oak throughout, and while very plain, presents an attractive interior. The total cost was about \$33,000. All the plumbing is "open."

The laboratories for the department of biology are so located as to use north light so far as possible. To that end, the room for museum purposes is on the south side of the building, as shown in the annexed floor plan. A similar museum room on the second floor is also devoted to the department of biology. These museum rooms are not large, and were not intended for display purposes, but mainly for the convenient storage of materials used in illustrating the lectures of the department. A lift runs through the entire height of the building, and by this it is easy to convey material from the second floor to the first, as it is required for use.

Between the museum and the lecture room is an apparatus room, or preparation room, which is used both in connection with the work of the museum and in preparing material for lecture work. The lecture room, in the southeast corner of the building, has the floor raised in four steps, so that all students can see the lecture table with clearness. The blackboard is a sliding one, and back of it is a fixed plate of ground glass which can be used for illustrative purposes in the lecture, or can be used in connection with the lantern work. The windows are fitted with opaque shades, so arranged as to exclude the light, in order that the room may be used for lantern work.

Adjoining the lecture room, on the north, is the bacteriology room. This is intended as a place where the various forms of apparatus connected with the work in bacteriology can be permanently set up. The room is large enough, also, so that small special classes in bacteriology can do their work there.

The two rooms at the west end of the building are the general laboratories, used for the more elementary work, and for the vertebrate work. They are so arranged that by a movable screen the two rooms can be used separately or as one large room for especially large classes.



At the right of the north entrance is the private laboratory and office of the head of the department. Adjoining this on the west is another private laboratory, from which a door opens immediately into the general laboratory. At the left of the north entrance is a room used for the more advanced classes in microscopical work. This opens by double doors into the laboratory in the northeast corner of the building, which is also used for microscopical work. By means of the double doors these two rooms can be thrown together, in case of exceptionally large classes. Inasmuch as the building is seldom used in the evening, and as it was necessary to have gas in all parts of the building in any case, the building has not been wired for electric light.



PRIVATE LABORATORY AND OFFICE.

In the laboratories for microscopical work, each table is supplied with gas jets, and when it is necessary to use artificial light for class work, portable gas lamps with Welsbach burners are furnished the students. In the author's experience, the Welsbach burner is by far the most pleasant light for microscopical work. The plans show clearly the position of the sinks and the acid closets, which are distributed freely in the various laboratories. In the arrangement of the tables all are so placed as to face the windows. The author has a personal objection to microscopical work with a side light. It is to him extremely annoying, and it seems desirable that the student should work under the best conditions; consequently, in the various laboratories, the tables are arranged in two lines, one on the wall immediately facing the windows, and the other a few feet back. It is found that this gives much better working light than to arrange the tables in alcove fashion from the walls, and it is also fully as economical of floor space. Our room is so ample at present that it is possible in all our classes to

furnish students with tables which become their permanent property for the term's work. This has a great many advantages, as it is not necessary to clear the table after every work period, and it is possible, in the case of enthusiastic students, to put in more than their required time upon their laboratory work. As a matter of fact, in many cases, more than double the amount of required time is spent by many of the students in the laboratory.

For laboratory tables, after a good deal of thought, we have used hard wood kitchen tables. The essential points in a laboratory table for an under-graduate student seems to be a sufficient amount of room, and stability. These tables are well made—each has a drawer—and have the virtue of cheapness, and we prefer to spend our money in other forms of apparatus rather than in elaborate laboratory tables. The tops of the tables are painted a dull black.



ELEMENTARY LABORATORY.

For laboratory chairs, we have used a form which we have seen in no other laboratory. Many of our best laboratories supply stools for their students. It has always seemed to us rather hard that the student, who is working two hours or more continuously at the laboratory table, should have nothing more comfortable than a stool without a back. It is very desirable that whatever chairs are used should be adjustable in height, because of the varying heights of the students as well as the requirements of the different kinds of work. We have finally adopted in all of our biological laboratories a chair which is easily understood from the illustrations. The base is a swivel stool, upon which is placed the seat of an ordinary kitchen chair. These chairs are very comfortable, and at the same time very durable, and we have found them extremely satisfactory for the laboratory work.

In both the general laboratory and the histological laboratory, is a large drip tank (one of them is shown in the illustration of the elementary laboratory) so arranged that jars of living material can be kept alive by means of tubes from the overhanging water pipe, or the tank can be used as a large fish tank by plugging the outlet with a hollow plug of slightly less depth than the tank itself. In the general laboratory this tank is used for keeping such animals as fresh water mussels at the times when the classes are using them.

For storage of the thousand and one things that are in constant use in the laboratories, as well as the working collections in homeopathic vials and similar small receptacles, we have a series of tray cases (one of which is shown in the picture of the private laboratory). The trays are made of uniform size, and are



HISTOLOGICAL LABORATORIES.

interchangeable in position, or in the different cases. They run on one-half inch cleats on the sides of the cases, and each one has a label holder upon the front. In the trays that are used for homeopathic vials, partitions are inserted parallel with the front and back of the trays. For such collections we use the ordinary eight-drachm homeopathic vials. Arranged in this way, these trays will hold 195 vials each, thus being extremely economical of room. Where the trays are used for miscellaneous material, they are arranged alphabetically, so that it is easy to place one's hand upon corks of various sizes, bottles, etc. For the collections in homeopathic vials an accession catalog is kept, and the bottles are arranged according to the numbers of this catalog. By means of a subject catalog upon cards, it is easy to find any desired material. The trays are of such a size that they hold a definite number of Pillsbury slide boxes, so that they can be used also for packing away slide collections. The collection

of slides is also arranged numerically, the accession catalog being the one devised by Dr. Ward. The subject card catalog makes it comparatively easy to locate the slides of any particular subject.

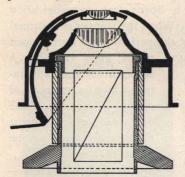
Between the museum and the general laboratory is a room in which is placed the department library, where are kept all the working books on biology. To this the students of the department have free access at all times.

The storerooms of the department are located upon the north side of the basement. Upon the south side of the basement are two rooms for botanical laboratories. On the south side of the basement, also, is located the vivarium, in which are kept the animals which must be kept in stock for the biological laboratory. This room is floored with cement, inclining towards an opening in the center which is connected with the sewer, so that by means of a hose the room can be thoroughly flushed and cleansed. Connected with this room is the injection room, which has a cement floor arranged in the same manner. In this room all the dirty work of killing large animals and of injection, in fact all work which would be liable to foul the laboratories, is taken care of. In this way a large part of the disagreeable work is kept out of the laboratory rooms.

Ripon College. C. Dwight Marsh.

## A Combined Condenser and Polarizer for Petrographical Microscopes.

The attachment consists of a double lens condensing system, and a Nicol prism mounted as shown in the following illustration.



The upper condensing lens is mounted on a revolving arm so that it may, at the will of the operator, be instantly thrown in or out of the optical axis by a lever; a suitable stop being provided to bring it to a central position.

The lower lens is mounted at the proper distance below the upper surface of the apparatus so that when the upper lens is moved out of optical axis, the lower lens focuses upon the slide, thus avoiding the necessity of refocusing the condenser system when changing

from the double to single combination. The Nicol prism is mounted in revolving sleeve with graduated collar and a stop to indicate zero or the position of Nicol prisms when crossed.

The advantages of this arrangement over others for accomplishing the same results are, briefly, as follows:

First—It is not necessary to increase the size or thickness of the microscope stage.

Second—The attachment is always in focus whether one or both lenses are used.

Third—Compactness and freedom from liability of disturbance while operating stage or slide.

W. L. Patterson.

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30c.c

#### A Plan for a Ureometer.

The plan for a ureometer (or modification of Doremus' apparatus) here presented would, it is believed, offer some advantages as to accuracy and facility of manipulation over other methods of estimating urea.

C is a tube of about 25 cubic centimeters capacity, closed at the upper end, and accurately graduated from the upper end downward for 16 c. c., to tenths of a cubic centimeter. The graduation also gives urea percentages to tenths per

cent., at 20°C, and barometric pressure of 760 mm. of mercury. At the bottom of tube c is a curved neck communicating with a bulb b, which opens into a tube a; this has a funnel top, and is of the same length as tube c. The capacity of bulb b is about 30 c. c., which quantity is indicated by a mark on tube a.

Tube c communicates near its lower end with a small tube d, joined at such an angle and position that gas generated in d will rise easily into tube c and none pass into bulb b. A removable accurately fitted glass stopper and stop-cock e separates tube d from the cavity of tube c, and the capacity of tube d below the stopper is exactly 1 cubic centimeter when the stopper is inserted. A transverse perforation n, rather large, passes through the stopper, so that when turned in the right direction it opens free communication between tubes d and c. When the stopper is so turned that it shuts off tube d, the perforation n should open into the cavity of tube c. A glass cross-bar h

strengthens the apparatus, which is entirely of glass. A separate base or stand for the support of the instrument can be provided.

METHOD OF USE. — The stopper, being removed, the tube d is by means of a dropper filled with urine. The stopper e is then inserted with opening n so placed as

to cut off tube d from tube c. Tube d then contains exactly 1 c. c. of urine. The usual sodium-hypobromite solution is then poured into tube  $a_t$  up to the 30 c. c. mark. The apparatus is then tilted so that the solution runs into tube c, entirely filling it; the perforation n should also be filled with the solution. The apparatus being held upright, the stopper e is turned so as to

make communication between tubes d and c. Tube d and perforation n should be large enough to enable the fluids to mix readily. The rapidity with which the fluids mix can be controlled by the stop-cock. The urine mixes with the test solution, the urea is decomposed, and the nitrogen evolved rises into the upper part of tube c. When the reaction is complete and the temperature has subsided to that of the room, water is added to (or removed from) tube a until the level of the fluid in the two arms is the same. The amount of gas in tube c is then read off, and from it the amount of urea can be calculated; or the percentage of urea can be read directly from the graduation.

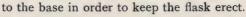
The advantages of this method would be as follows: The measured amount of urine (1 c. c.) is obtained accurately and easily, as it were automatically. None of the gas generated is lost, but all is saved in tube  $\epsilon$  for measurement. The equalization of the level of the fluid in the two arms equalizes the hydrostatic pressure and thus gives an accurate reading of the amount of gas free from that source of error. The graduation in cubic centimeters enables the urea to be calculated with the greatest nicety, applying any corrections for temperature and barometric pressure; while for localities near the sea level and ordinary room temperatures the percentage graduation on the tube (in the sketch given on the theoretical basis) gives a direct reading of sufficient accuracy. The apparatus is compact, not cumbersome, easily kept clean, and always in working order. It combines accuracy with facility of manipulation.

J. B. NICHOLS, M. D.

Washington, D. C.

#### A Device for Supporting Pasteur Flasks.

Pasteur flasks are difficult to handle on account of their peculiar shape. A collar of asbestos, cork, or straw is ordinarily used, but has to be fitted closely





The photograph shows a device for supporting these flasks, which permits greater freedom and safety in manipulation than is obtained with the ordinary collar support. The device consists of a solid disk of wood about  $5\frac{1}{2}$  inches in diameter and 2 inches in thickness. This is hollowed out in the center, leaving a concavity into which the base of the flask fits. One end of a piece of heavy brass wire is fastened into the margin of the base, the other end of the wire is bent so that the bend of the tube of the flask fits into it loosely. The wire supports the flask in the erect position, so that the base of the flask need not fit closely into the hollowed wooden base.

#### LABORATORY PHOTOGRAPHY.

#### HIGH-POWER PHOTO-MICROGRAPHY.

There is a fascination about the use of the microscope and camera together that can hardly be experienced when either instrument is operated alone. In its simpler aspects, moreover, photo-micrography may be enjoyed by any one who possesses the ordinary microscopical and photographic apparatus. By makeshift adjustments and adaptations, it is possible to arrange the separate parts into a workable series so that the amateur photographer may add the making of enlarged pictures of small objects to his other accomplishments and the microscopist may secure permanent records of the transitory images that have so often delighted him.

It is otherwise, however, with those who attack the problem of producing photo-micrographs which represent a high amplification of the object—1000 diameters or over. This branch of the work should not be undertaken without serious purpose and the best of apparatus.

Here makeshifts are out of place. The great degree of accuracy and the nicety of adjustment demanded of each part of the apparatus makes it necessary to employ an installation that is especially designed for its own particular purpose. With such assistance, only, can the scientist achieve any valuable results, for it is only the scientist who would have the time and patience requisite for work of this character. There must be some end in view aside from the mere gratification of an idle curiosity to see how big a picture of a small object can be made. This purpose finds itself in the desire of investigators to present to their fellows as accurate and as complete a conception of their material as it is possible to give. The value of photography as an aid in this direction is being more and more appreciated and nowhere more than among those who have to deal with the almost ultra-microscopic structure of the organic cell.

Not that photographs are designed to supercede the customary drawings. Both sun image and pencil image have their places as aids in the elucidation of the text. The former exhibits, often in a bewilderment of detail, the whole field of the study; the latter presents concretely the investigator's interpretation of the essential facts. A comparison of the two by one acquainted with the subject will enable him to reach an opinion as to the validity of the writer's conclusions such as would be impossible if only one method of delineation had been used.

In recognition of this fact, many writers upon histological and cytological subjects now enrich their papers by supplementary plates of photographs and drawings, which, with the text, enable a reader to obtain as complete a mental image of the subject as can be acquired without a personal examination of the specimens. Such a work as Wilson's "Fertilization and Karyokinesis of the Ovum," wherein the author's skill in observation is supplemented by the beautiful photographs of Dr. Leaming, is an excellent example of what may be done in this direction.

As a realization of the importance of this class of illustration grows, the

demand for information concerning the methods employed increases until microscopical journals find it advantageous to assign a separate department to the discussion of methods and apparatus involved in the production of scientific photographs. It is a matter of congratulation that workers in this country are now afforded such a means of communication through the columns of the Journal of Applied Microscopy and Laboratory Methods.

The department here being new, it is but natural that matters of an elementary nature should be discussed. With this in mind, I have thought to describe an actual installation of apparatus for high-power photo-micrographic work and to exhibit some of the results attained by its use. In Vol. III, No. 5, of the JOURNAL appeared an account of the outfit employed in the Johns Hopkins laboratory. The one at the University of Kansas is very similar, but it has been



FIGURE 1.—Spermatogonial mitoses of the grasshopper, Hippiscus phænicopterus, in the metaphase and anaphase. These divisions take place very rapidly and the archoplasmic threads of previous spindles may still be seen between the centrosomes of different cells. 1000 diameters.

further modified from the original Zeiss arrangement than has the one at Baltimore. Upon the optical bench are placed the illuminating apparatus, two iris diaphragm supports, and the microscope. The other accessories furnished with the complete outfit are not employed. The camera itself has not been altered.

Following is the arrangement of the apparatus: The microscope—I use a Van Heurck—Watson stand—is firmly clamped on the end of the bench nearest the camera. Next, the carbons of the arc light are roughly adjusted so as to lie approximately within the optical axis of the microscope. With a low power objective focused upon the object, the arc is projected upon a small screen suspended upon the front of the camera which is pushed back on its sliding bed to a distance. By means of the adjustment screws, the arc is then brought into such a position that the glowing crater occupies the center of the field. Prelim-

inary to this, however, the substage condenser has been racked up with the coarse adjustment until it brings the image of the crater into focus at the level of the object. If the camera has not been adjusted with reference to the optical bench, it is now arranged so that the image of the crater falls in the center of the ground glass. Provided the substage condenser is properly centered, the linear adjustment of the combination is complete.

The next step is to arrange the object with reference to the condenser and the objective which is to be used in making the negative. I have found it advantageous to connect these three elements with homogeneous immersion fluid and for a condenser employ the "parachromatic" oil immersion form made by Watson & Sons. The objective is an apochromatic 2 mm. of Zeiss.

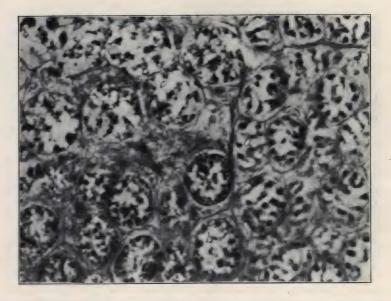


FIGURE 2.—Transformation stages between the telophase of the spermatogonia and the prophase of the first spermatocytes. The gradual accumulation of the chromatin into a thread may be noted.

Successive stages shown at "a", "b", and "c". Same object. 1000 diameters.

For a number of reasons, it is convenient to interpose temporarily an incandescent gas lamp in the substage series while getting the proper focus and adjustment of the object with the eye. When this adjustment has been accomplished, it remains only to get the final projection of the image upon the ground glass of the camera before the exposure is made.

The projection eyepiece is now substituted for the observation ocular, which has been used up to this time, and an image thrown upon the small screen which still hangs upon the front of the camera. Here an approximate focus of both object and source of illumination is obtained and the composition of the picture studied. If this is satisfactory, the screen is removed, the camera pulled forward and joined to the microscope, and connections made between the fine adjustments of the tube and of the substage condenser with rods that lead back to the

end of the camera carriage. Here these may be manipulated while the image is being examined and focused on the screen.

At this stage of the proceedings, there are visible upon the ground glass indistinct images of the incandescent carbon and of the object. By means of the fine adjustments, these are brought into a sharp focus upon the glass. Owing to the nature of the crater, the illumination is not uniform over the whole field and it is necessary to place a piece of ground glass between the source of illumination and the object. This should not be more than dense enough to properly diffuse the light, otherwise it unduly lengthens the time of exposure.

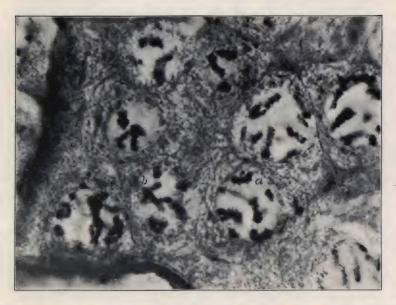


FIGURE 3.—Prophase of the first spermatocyte. The chromatin thread has broken into segments. Between these run delicate linin fibers as at "a". At "b" an element distinguishable from the others by its greater transparency and sharper outline. In these cells it is known as the accessory chromosome. Same object. 1000 diameters.

There should now be visible a sharply defined, evenly illuminated image of the object wherein the details are neither obliterated by excessive illumination, nor rendered granular and obscure by deficient light.

In obtaining a good negative, the manipulation of the substage iris diaphragm is as important as the proper adjustment of the objective. No pains are spared, accordingly, to bring about the appropriate arrangement of the cone of light, and the lever of the diaphragm is swung back and forth until the very best possible result is obtained. The Watson condenser above mentioned has a graduation upon the mounting indicating the numerical aperture afforded by the opening of the diaphragm. This is a convenience which should find a place upon the condensers of other manufacturers.

The further stages of the process are those which pertain to photographic manipulations in general, and the limits of this article will not permit their consideration. Summing up the steps to be followed, we have:

- 1. Linear adjustment of substage condenser, crater, and center of cameraback to obtain their coincidence with the optical axis of the microscope.
- 2. Focus of source of illumination upon object by means of substage condenser.
  - 3. Focus of objective upon object.

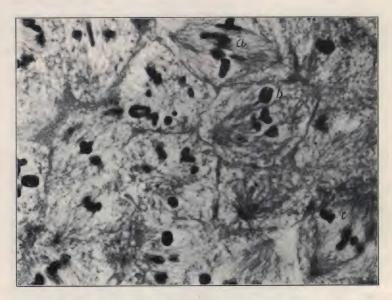


FIGURE 4.—Metaphase of the first spermatocyte. The chromosomes of the same cell do not divide simultaneously, as may be seen at "a". Sometimes they form rings as seen at "b".

In the cell marked "c" the archoplasmic fibers are sharply in focus. Same object. 1000 diameters.

- 4. Final simultaneous projection of crater and object images upon ground glass of camera.
  - 5. Diffusion of light by ground glass between source of illumination and object.
  - 6. Adjustment of substage iris diaphragm.
  - 7. Exposure of plate.

C. E. McClung.

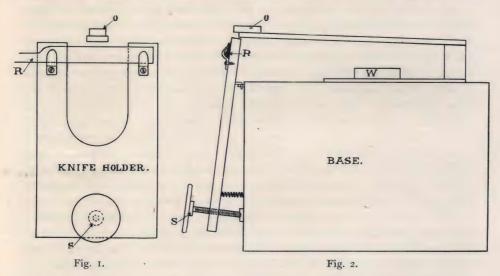
University of Kansas.

#### An Improvised Microtome.

On a recent visit to the College of Physicians and Surgeons in Boston, the writer saw, among other ingenious contrivances, a microtome devised by Dr. Shurtleff, of the above named institution, and made by him at a cost of fifty cents, for cutting the micrometer screw. Since my visit I have myself made a similar microtome at the small cost of two hours' work, since a common screw was employed instead of a micrometer. Thinking that possibly the idea may be of use to some other investigator, I will venture to offer the following description:

The first essential is, of course, a knife; and, while a regular section razor is preferable, an ordinary razor will answer. In the absence of either, however, I

have seen a shoe knife successfully used; but in this case the back was strengthened by soldering a knitting needle on one side and a rod cut from a stove poker on the other. Assuming, then, that a knife is at hand, the next requisite is the holder, which consists of a piece of wood about four by seven inches, having a U-shaped cut-out at the top, two inches wide and three inches deep. This leaves two prongs each an inch wide into which small wire nails are driven so that the razor (R) may rest upon them when it is in position. Spring clips of some sort hold the razor firmly in place. For the clips, stout wire an eighth of an inch thick is good. Each wire may be fastened to the board by double-pointed tacks. Near the bottom of the board and in the center is the screw (S). A common screw will answer, but a fine threaded screw passing through a nut is better. In either case, however, a large disc may be soldered to the screw head for increased delicacy in operation. The "holder" complete is now, by means of a pair of hooks and eyes, to be made attachable to the end of a box so that turning the screw



gives a delicate movement to the razor. The screw point should work against a small metal plate on the box. Tension is secured with a rubber band or spiral spring. (Reference to the diagram will make the idea clear.) The "holder" should be so placed that the razor edge will be two or more inches higher than the top of the box. Now, when an adjustable object-holder is provided, the microtome is completed. To make the object-holder, a board somewhat shorter than the box, a block, and a straight-grained stick about one-half an inch in cross section are necessary. Fasten the block near one end of the board, nail the stick to the block (as indicated in the diagram), and the microtome is ready for service.

In use, the paraffin block (O) is fastened to the end of the stick with melted paraffin, and proper adjustments are made with reference to the razor. Then, downward pressure on the stick cuts the section, while clockwise movement of the screw regulates the thickness. Serial sections are readily made, if the

paraffin block is carefully squared; but, for this work, the object-holder should be steadied by a weight of five or six pounds (W).

While the microtome can by no means take the place of such a splendid instrument as the Bausch & Lomb "Student," yet it is a practical and serviceable apparatus, and its usefulness has been demonstrated in everyday histological work.

IRWIN LAVERNE POWERS.

Randolph, Mass.

#### The Study of Bacteria in the Public Schools.

The highest aims in "municipal housekeeping" can never be attained by Boards of Health or by Departments of Street Cleaning alone, however efficient these organizations may be. Unless these city departments are backed by a strong, intelligent public sentiment we shall experience nothing better than sporadic reform in the cleaning of our streets, in the construction of tenement houses, and in the general care for the public health. When conditions get sufficiently bad in a community, it is comparatively easy to arouse the voters and roll in a reform administration by big majorities. But alas! we soon tire of our attempts at public virtue, we reverse our votes at the next election, and sink back into easy toleration of filth and its resulting disease. One might indeed become pessimistic with reference to the future of our cities were it not true that democracy possesses a most powerful means of developing a public sentiment which may be at once intelligent and lasting. Gathered in our schools of to-day are the boys and girls who will be the voters and the home-makers of to-morrow. Hence to the teacher, especially in the public schools, is given the opportunity to exert a telling influence in developing the better city of the future.

The discoveries in bacteriology within a few years have made new sciences of surgery, medicine, and sanitation. Epidemics of typhoid fever have ceased to be regarded as "a dispensation of an all-wise Providence," for we have come to know that the presence of this disease usually means a contaminated water supply or imperfect sewerage. Scientific men have learned, too, how to check the ravages of yellow fever and cholera, and even consumption is found to be a preventable disease. To make these discoveries of practical use, however, this knowledge must be possessed by a large majority of the citizens in a community, and the most effective means of attaining this end is by educating the pupils in our public schools. With this object in view, in the Peter Cooper High School, New York City, we devote considerable time in the course in biology to the study of bacteria, yeast, and moulds.

In this study, it is necessary at the very first to impress the pupil with some idea of the omnipresence of these micro-organisms in everyday life; and for this purpose an experiment performed by the boy or the girl is always more telling than a talk by the teacher or a dozen pages of description. We begin with the study of a hay infusion. The work is done by each pupil at home, and the report presented at the next recitation. The following account is selected from the one hundred and fifty papers received from the first year pupils:

"Straw Infusion. I procured about a handful of straw at a livery stable and put it in a Mason jar three-quarters full of water, and put it in a warm place where the temperature was on an average of 75°, on Thursday, March 22. Its color was tan and the mixture smelt like musty straw.

"The 23d, temperature 73°, mixture getting darker in color, and smell becoming more noticeable. Saturday, temperature 74°. A thin scum is forming and small things are coming up from the bottom and straw. The smell is getting very strong.

"Sunday, temperature  $74^{\circ}$ , scum becoming thicker and bubbles appearing in it." Discussion and microscopical examination in the class room brought out the fact that the scum was composed of countless bacteria and other micro-organisms which had grown from the germs on the dried hay. The inference was drawn from the experiment that bacteria grow rapidly in a warm temperature, when water and organic matter are present, and that decay is one of the results of their activity.

The cultivation of bacteria in the laboratory was the topic next considered. Nutrient gelatin, the most useful medium in which to grow all kinds of bacteria, may be readily prepared in the laboratory or in the home kitchen. The ingredients necessary are the following: one pound of lean beef chopped fine (or better run through a meat cutter); 60 grams (2 oz.) of the best French gelatin; 6 grams (1-5 oz.) of peptone, which can be bought for 10 cents at any drug store; a teaspoonful of salt, and a little baking soda. Put the beef in a porcelain or agate dish, add a pint of cold water, and allow the mixture to boil slowly for a half hour. Strain the broth through muslin and then allow the liquid to run through filter paper. Pour in enough water to make the quantity of broth equal to about a pint and a half.\* The gelatin, cut into small pieces, is then added to the broth, together with the peptone and salt. The mixture should be heated sufficiently to cause the gelatin to dissolve, but should not be allowed to boil. Just enough cooking soda is added to cause red litmus paper dipped in the mixture to turn blue, that is, the liquid should be faintly alkaline. Filtering the hot gelatin sometimes involves more or less difficulty. The process can be easily carried on, however, within a steam cooker. A glass funnel should be put in the mouth of a Florence flask (used commonly in a chemical laboratory) and one or two layers of absorbent cotton placed within the funnel. If the gelatin, flask, and funnel are kept hot within the cooker the liquid will readily pass through the cotton. After filtering, close the mouth of the flask with a plug of absorbent cotton, and boil for a few moments. The flask may be set aside as stock gelatin until needed for use. (If the gelatin mixture is not clear, it should be filtered through the same cotton a second time.)

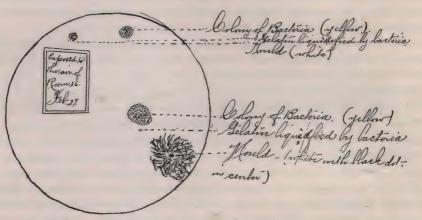
Some of the liquid gelatin was poured into clean Petri dishes (Fig. 1), or test tubes plugged with cotton may be used. After the gelatin had solidified some of the dishes were opened to the air. Several days after this exposure the cultures were placed upon the desks of the pupils, and they were asked to make drawings of the bacteria colonies, and to answer certain

<sup>\*</sup>This broth may be prepared more easily from Liebig's beef extract. Four grams should be dissolved in the pint-and-a-half (750 c. c.) of water, and the solution should be filtered through filter paper.



Fig. 1. Petro dish with cover.

David S. Kelly J. 2.
The Study of the Bacteria.



The dish that has not been exposed to the air is perfectly clean but the dish that has been exposed have colonies of bacteria and moulds We shudied these dishes on the first of March when the colonies appeared like little yellow dots on Harch a we studied these same dishes and the above diaming shows the development of the colonies The gelatin around these colonies has been liquided by the action of the lacteria informit."

Fig. 2.

questions stated in the Laboratory Manual. One of the papers prepared during a recitation period is reproduced in Fig. 2. Figures 3 and 4 are drawings of Petri dish cultures made by two other pupils.

One of the boys, not satisfied with the amount of laboratory work given in school, prepared nutrient gelatin at home. He writes thus of his experiences:

"I took about a half pound of lean beef and after cutting into pieces placed in a pot and covered with water, then brought to a boil. I should also mention that I used a moderate fire so that the process occupied about twenty minutes. After obtaining my broth I added gelatin and brought again to a boil. Here I added some salt and carbonate of soda, after which I strained the broth through cotton into a sterilized bottle and corked.

"I experienced such trouble in clearing the gelatin of colonies that I finally melted the gelatin and poured it into test tubes and in them brought it to a boil with the result of one tube burnt and five cleared. In three of the tubes Mr. Peabody inoculated pure cultures; one of the tubes has produced a very large red colony, the others have not grown."

This laboratory work on the growth of bacteria was followed by an experiment performed at home by the pupils. One of the girls gives the following report of her work:

"The Study of Bacteria in Milk. I procured three bottles of about the same size. I then thoroughly cleansed each bottle before I used it. Two of the bottles had stoppers; the other had none. One of the bottles I half filled with good fresh milk, put the stopper on, and set it outside the window. I labeled this bottle 'No. 1.'

"Into the second bottle I poured about the same amount of milk, and set it aside in a warm temperature of about 70°. I labeled it 'No. 2.'

"The third bottle I cleaned in very hot water. I then boiled the same amount of milk that I put in each of the other bottles. I allowed it to boil for about three minutes. After the milk had cooled a little I poured it into the third bottle. I placed it beside bottle No. 2, and labeled it 'Sterilized Milk.'

"At the end of fifteen hours I examined each of the bottles. I noticed that No. 1 had very little smell at all. No. 2 had a sour like smell. It smelled as if the milk were turning. No. 3 had hardly any smell at all. If there was any smell at all, it was a sweet one. I now boiled the milk in No. 3 again. I first thoroughly cleansed the bottle and cork before I put the milk in. I then placed it beside No. 2, and put No. 1 again outside the window.

"At the end of twenty-four hours I again examined my bottles. I found that No. 1 had not any smell at all. No. 2 had a very decidedly sour smell, and No. 3 had a sweet smell.

"The changes in the milk are due to the growth of the bacteria from the air, or on the bottles, or the stoppers. As far as my experiment has worked I do not think a cold temperature kills the bacteria, but I think it numbs them. I think a boiling temperature kills the bacteria, and I think a moderate temperature increases the growth of the bacteria."

Successful microscopical work was done with magnifying powers of about 500 diameters. Pure cultures of spherical-, rod-, and spiral-shaped bacteria growing in test tubes of gelatin were supplied us by Dr. T. Mitchell Prudden of the

College of Physicians and Surgeons, to whom I am much indebted for help in this bacteriological work. Microscopical slides are easily prepared thus: Hold upside down the test tube in which bacteria are growing, and carefully remove the cotton from the mouth. Touch one of the colonies of bacteria with the point of a needle, and then rub the needle point on a clean glass slide; add a drop of water to the spot touched by the needle, cover with a cover-glass. Stains (Loef-fler's methylen blue and Ziehl's carbol fuchsin) bring out more clearly the structure of the bacteria. Each of the thirty-five pupils in a division examined the stained bacteria, and watched under another microscope the motion of the living forms. One pupil's written account of this study is here given:

"MICROSCOPIC STUDY OF BACTERIA. 1. The bacteria which I saw under the microscope last Wednesday were of red and blue colors. This was caused by the coloring matter (stains).

- "2. They were of three different shapes, round, pencil-shaped, and corkscrew.
- "3. The bacteria which I saw to-day under the microscope are moving around.
- "4. There were also under the microscope egg-shaped animals which were moving around." (This slide was prepared from the hay infusion and contained infusoria.)

A little mathematical problem worked out by each student helped to make real the rapidity of multiplication among these micro-organisms. The pupils were told that a rod-shaped bacterium, when conditions are favorable, divides in about an hour to form two bacteria. The problem was stated something like this: Suppose we start with a single bacterium this morning at 10 o'clock; if conditions are favorable, how many cells would be seen at 11 o'clock? The answer was "two." Between 11 and 12 o'clock each of the two would divide to form two; hence at 12 o'clock it was evident that there would be four bacteria in place of the single cell at 10 o'clock. The pupils, continuing the calculation, found that if the process were to go on until 10 o'clock the next morning, the original bacterium would give rise to 16,776,216. The completion of this calculation for a second day's crop of bacteria was not attempted for obvious reasons.

Thus far the experiments and discussions had made real to the pupils the existence of countless millions of micro-organisms. They had learned something of the form, size, and motions of the individual bacteria; and they had become acquainted with some of the results of their activity in causing decay, in souring milk, and in producing colors.

Some of the conditions which tend to check the growth of bacteria were learned from the milk experiment performed at home. A laboratory demonstration developed this subject still further. One of the boys described the experiment thus:

"STERILIZATION. Mr. Peabody took three test tubes and inoculated some of the bacteria from the hay infusion. The first test tube contained nourishment in a solid form (nutrient gelatin), and after the bacteria had been inoculated it was set aside. The second test tube was prepared the same way, but Mr. Peabody poured some corrosive sublimate over the surface of the gelatin. The third test tube was prepared in the same way as the first, but was put in the (steam) sterilizer for five minutes, and then set aside.

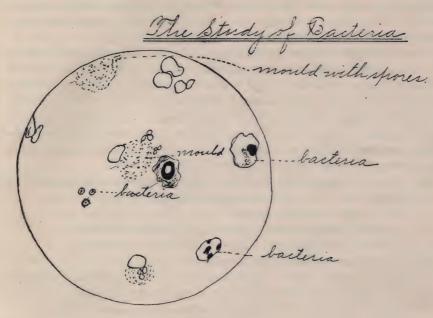
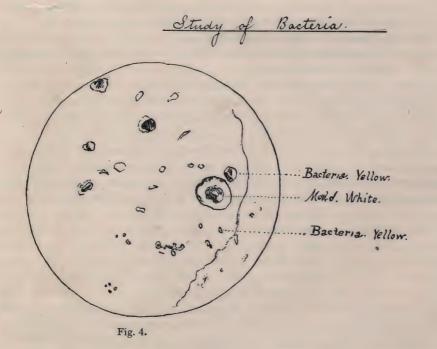


Fig. 3.



"At the end of five days we examined the tubes and found that the two tubes, one sterilized by heat and the other by poison, were perfectly clean, while the other had a large colony growing. From this I infer that corrosive sublimate and the heat killed the bacteria."

We are fortunate in possessing a hundred copies of "The Story of Bacteria" and a like number of "Dust and its Dangers" by Dr. T. M. Prudden. These books were loaned to the 192 pupils who were studying the subject, and about one-fourth of the chapters were assigned for text-book lessons. One may judge of the interest in this study by the following figures: When the books were returned it was found that 103 pupils had read the whole book; that the books had been read by 197 parents or friends of the pupils; and that various topics in bacteriology had been discussed in over half of the homes.

The practical applications of the subject were brought out in discussion of a list of questions from which the following are selected:

- 1. From all your experiments state
  - a. What conditions seem to favor the growth of bacteria?
  - b. What conditions seem to hinder the growth of bacteria?
- 2. Why are fruits cooked before canning?
- 3. Why should fruit jars be filled completely before screwing on the cover?
- 4. Why is grass dried before putting it in the barn?
- 5. Why are milk, meat, etc., put in the refrigerator in summer time?
- 6. Why should the prohibition against spitting in public places be rigidly enforced?
- 7. Why should sweeping be done as far as possible without raising a dust?
- 8. Why are hard wood floors more healthful than carpets?
- 9. Why should the teeth be brushed often?
- 10. Why should the refuse be removed from the streets every morning early, especially in summer time?
- 11. Why should sink drains be carefully inspected?
- 12. Why should wounds be carefully cleansed and dressed at once?
- 13. Why are typhoid fever, diphtheria, and other infectious diseases often best treated in hospitals?

The tables of the New York Board of Health give figures and charts which serve to clinch the argument in favor of good city housekeeping. The pupils copied into their note-books the following figures giving the annual death-rate per thousand of the population in New York City, 1886 to 1896 inclusive:

1886, 25.99	1891, 26.31
1887, 26.32	1892, 25.95
1888, 26.39	1893, 25.30
1889, 25.32	1894, 22.76
1890, 24.87	1895, 23.11
1896, 21.52	(first part of year).

There was little need to suggest that the sudden decrease in death-rate in 1894 and in succeeding years was doubtless due in no small measure to the efficiency of the Street Cleaning Department organized and directed by the late Col. Waring.

After reading Dr. Prudden's books, and after class-room discussions, each pupil was asked to outline at home the arguments in favor of and against the bacteria. The case is stated thus in one of the papers:

- "BENEFITS OF BACTERIA TO MANKIND. They construct food-stuffs for plants out of the nitrogen gas and the solutions absorbed from the soil.
  - "They ripen the cream before churning and thus form butter.
  - "They give flavor to butter.
  - "They are an absolute necessity in making cheese.
  - "In making vinegar from cider, yeast and bacteria work together.
- "Bacteria perform a very necessary work in the process of 'retting' flax in the linen industry, without which we would not have our fine linen and delicate laces.
  - "Bacteria play a prominent part in the curing of tobacco.
  - "Sprouting of seeds is promoted by bacteria.
  - "Streams and lakes are cleared by bacteria.
  - "They decompose dead animals into the dust from whence they came.
- "The Ways Bacteria Prove to be 'Man's Invisible Foes.' Bacteria cause the diseases, consumption, typhoid fever, scarlet fever, pneumonia, leprosy, lock-jaw, influenza, cholera.
  - "They cause blood poisoning.
  - "They destroy foods."

The primary aim of these eight lessons in bacteriology, as already stated, was a practical one, namely, to present to the boys and girls of our city a most telling argument for cleanliness in the care of the home and in the care of the city. The colored charts portraying the cases of consumption in the region of Mott street and of diphtheria in the Tenth and Twelfth wards will not soon be forgotten. Hence the New York of to-morrow will doubtless number among its citizens at least a few more staunch supporters of an efficient Board of Health; a few more homes will probably be free from the danger of disease contagion, and a few more house-wives will exercise greater care to secure abundance of light and of fresh air in their homes and to select and prepare nutritious foods.

The treatment of the subject, however, was not allowed to leave in the minds of the pupils the lasting impression that we have discovered in bacteria an omnipresent and well-nigh omnipotent enemy. They were led to see that consumption, cholera, typhoid, and all the other diseases charged to these micro-organisms are due to the ignorance or carelessness of man, and that these diseases can be prevented. While, on the other hand, they learned that the bacteria are toiling incessantly to clear our earth from the debris of decay, and to prepare the soil and the air for the growth of the higher plants. Thus this study becomes a part of the great study of biology, and in this fact lies the deeper interest of the subject. In the hay infusion all the functions of living nature are in full operation. There one may study assimilation, oxidation, respiration, excretion, the life and death struggle for food, reproduction, and even something akin to sensation; for who of us, after an hour at the microscope, watching the varying movements in this world of micro-organisms, is prepared to deny absolutely all sentient impressions even among bacteria? Biological study of this sort should

not only result in more healthy bodies for our pupils and in a more healthful community, but it should contribute largely to broaden and deepen the mental life of the student.

James E. Peabody.

The Peter Cooper High School, New York City.

#### Biology Wall Charts.

"A Method of Making Biology Wall-Charts," by F. D. Heald of Parsons College, Fairfield, Iowa, published in the JOURNAL OF APPLIED MICROSCOPY for November, 1900, induces me to speak of a method of chart making which I have adopted with considerable satisfaction, to myself at least.

The method is not original with me, but was suggested by Prof. H. P. Johnson of the University of California. My charts are made of material such as is used by "millers" for the manufacture of flour sacks. It is well known that this sack muslin has incorporated in the meshes of the cloth a filling of paste material which renders the surface smooth and very suitable to draw upon. Instead of a pen I use an ordinary paint brush of suitable size and shape. My pigments are such as painters use for the ordinary canvas advertising streamers and are procured ready mixed at the paint shop at a cost of a few cents. With these materials, charts of all sizes, colors, and kinds may be readily made. I find these "home-made" charts more satisfactory in my classes than any others that I have heretofore used, as there can be represented upon them exactly what it is wished to illustrate. These charts are so inexpensive and so easily made that any school may provide itself with a sufficient number for illustration in Physiology, Zoölogy, Botany, and other subjects. As they are made in watercolors, when not in use they should be kept in a dry place.

ORSON HOWARD.

University of Utah, Salt Lake City.

#### Staining in Toto with Delafield's Haematoxylin.

The stain is prepared according to the method found in Huber's "Directions for Work in Histological Laboratory," p. 153; except that, before using, it is diluted with an equal amount of distilled water, instead of five to ten times with water, as according to the directions.

The specimens, which should not be of too great size—not more than one-fourth inch in thickness—are left in the stain five days. After rinsing in water they are decolorized for two hours in acid alcohol made as follows:

Hydrochloric Acid, .		15 ·	1 part
Alcohol, 96 per cent.	•		 70 parts
Water,			30 parts

They are then washed in running water for at least two hours to remove all the acid. Dehydrate, and imbed in paraffin.

Newton Evans, M. D. American Medical Missionary College.

# Journal of Applied Microscopy

Laboratory Methods.

Edited by L. B. ELLIOTT.

Issued Monthly from the Publication Department of the Bausch & Lomb Optical Co., Rochester, N. Y.

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"Men of science form, as it were, an organized army, laboring on behalf of the whole nation, and generally under its direction and at its expense, to augment the stock of knowledge as may serve to promote industrial enterprise, to increase wealth, to adorn life, to improve political and social relations, and to further the moral development of individual citizens." The full significance of these words, written by Helmholz nearly half a century ago, is now only beginning to be fully appreciated. Scientific men, in spite of the old popular idea to the contrary, have

demonstrated indisputably their ability to cope with problems of the greatest practical and economic value. Industrial progress is more and more dependent upon the results of their labors.

The policy of our government has always been to support liberally men and institutions which undertake to promote the welfare of the people; and yet we must admit that we have not met our highest possibilities, for we have but to look to certain other progressive nations to see points wherein we can make decided improvement. It is no longer a theory that governmental support of scientific work pays in every sense of the word, for Germany has long since demonstrated it to be a fact. Her scientific instruments have been brought to the highest degree of perfection, by coöperation with individuals capable of improving them, and through them her industrial progress has been most advanced. "Made in Germany" has been a key to every market in the world. This development must be attributed to the coöperation of science and government; a condition of mutual support, toward which our own country is rapidly trending.

The work accomplished by our science departments and bureaux is only the preface of what may be expected in the near future. Nation, state, university, and individual are forming one great combination for the pursuit of pure and applied science. In this cooperation the needs of science will be largely brought to light by individuals who are actually engaged in the work. These needs must be met by the institution, state, or nation in whose interest the individual pursues his investigations.

There are now needs which handicap our progress and place us at a disadvantage in the competition with other nations. Governmental coöperation in the development of the methods of chemical glass, and many other manufactures, and in the standardizing and control of apparatus used for weighing and measuring, together with the adoption of the metric system of weights and measures, would produce practical results.

Scientific men have adopted the metric system in their work. The industries, recognizing its advantages, do not wait for the adoption of the system by the government, but are rapidly introducing it into their various calculations.

#### CURRENT BOTANICAL LITERATURE.

CHARLES J. CHAMBERLAIN.

Books for review and separates of papers on botanical subjects should be sent to Charles J. Chamberlain, University of Chicago, Chicago, Ill.

#### REVIEWS.

Juel, H. O. Beiträge zur Kentniss der Tetradenbildung. Jahrb. f. wiss. Bot. 35: 626– 659, pls. 15-16, 1900. This important contribution really consists of three distinct papers, which can be considered separately.

I. Tetrad formation in the ovule of Larix.

The homologies between the reproductive organs of the vascular cryptogams and the phanerogams have long been known in their grosser features. The pollen chambers in the anther are microsporangia and the pollen grains are microspores. The ovule is a modified megasporangium and the embryo-sac is a megaspore; but while it is accepted that the pollen grain, like the spore of a vascular cryptogam, arises by a tetrad division, it is generally believed that the embryo-sac is formed without a tetrad division.

Dr. Juel investigated the ovule of Larix sibirica from an early stage in the development of the mother cell of the megaspore up to the beginning of endosperm formation. The paper is of particular interest because it is the first to treat this portion of the life history of a Gymnosperm from the standpoint of modern cytology.

In material collected about the middle of April, before the snow had disappeared, the mother cell of the megaspore was easily distinguished by its large size and by the abundance of starch which it contained. The first division is heterotypic and shows the reduced number of chromosomes (12). At the poles of the spindle are granular masses which may possibly represent centrosomes. During the anaplase the starch disappears, a cell wall is formed and each of the daughter nuclei divides again by a homotypic division and thus gives rise to a row of four megaspores, the lowest of which germinates and produces the prothallium.

By comparing these series with the development of the microspore from the mother cell, which has already been thoroughly studied in *Larix*, Prof. Juel comes to the conclusion that the two series are homologous, the megaspore arising like the microspore by a tetrad division. While this conclusion is not new, the evidence supporting it is a real contribution.

#### II. The tetrad division in a hybrid plant.

It has long been known that hybrids are generally sterile, and it has also been known that the pollen of hybrid plants is commonly imperfect. The present writer investigated the formation of the tetrad in *Syringa rothomagensis*, a hybrid between *S. persica* and *S. vulgaris*. The form did not prove to be a favorable one for such a problem, because the pollen of both parents is poor, in *S. vulgaris* about 50 per cent. of the pollen grains appearing to be incapable of

functioning, and in S. persica normal pollen grains being quite rare. The latter form is almost as sterile as the hybrid.

In all three forms the development is normal up to the formation of the pollen mother cells. In the hybrid *S. rothomagensis* it was found that while most of the divisions in the pollen mother cells were mitotic, there were also numerous cases of amitotic division, and abnormalities in the chromatin and in the achromatic figure were frequent.

#### III. The development of the pollen grain of Carex.

As a rule the pollen mother cell of a flowering plant gives rise to four pollen grains, but it has been reported that in the Asclepiadaceæ and Cyperaceæ the mother cell gives rise to but one pollen grain.

A careful examination of *Carex acuta* gave the following results: The wall of the pollen mother cell becomes the wall of the pollen grain. The tetrad divisions take place, but the walls separating the four cells are imperfect and only one cell of the tetrad develops into a pollen grain, the other three being crowded out, just as in the megaspore series three potential megaspores are crowded out by the one functioning megaspore.

C. J. C.

Dixon, H. H. On the first mitosis of the spore mother cells of *Lilium*. Notes from the Botanical School of Trinity College, Dublin. No. 4, pp. 129-140, pls. 7-8, 1901.

The author states very clearly the points in regard to which there is essential agreement among cytologists,

and outlines the debated questions. While admitting that there is still ample room for dispute, he concludes that in both the first and second nucler divisions by which the spores are formed from the mother cell, the splitting of the chromosomes is longitudinal and that, consequently, there is no reducing division.

C. J. C.

Holm, Theodore. Erigenia bulbosa, Nutt. A Morphological and Anatomical Study. Am. Jour. Sci. IV. 11: 63-72. 6 figs. Mr. Holm, who has done more than any one else in this country on the minute anatomy of plants, presents in

his latest paper some interesting morphological and anatomical facts on this unique plant. The Erigenia possesses a single cotyledon. The blade of the cotyledon is held in a horizontal position and raised above the ground by a long slender petiole. In the second year after germination the first proper leaf appears and has a ternately decompound leaf, with divisions of the same shape as those of the mature leaf. The third year's growth is not much advanced as only a single green leaf is developed with a few additional divisions. The Erigenia germinates then with only one cotyledon.

The tuber as it appears during the seedling stage, as Holm has shown from anatomical considerations, is a swollen part of the primary root.

The structure in fully matured specimens is very different, and here is what the writer says concerning this:

"The mature tuberous root possesses a number of cork-layers, a secondary bark of very considerable width, filled with starch, and inside the bark is a band of collateral mestome-bundles with cambium between the leptome and hadrome and besides well defined strata of interfascicular cambium, while a broad pith occupies the central portion of the root, of which, however, the innermost part is broken down into a cavity; thus the principal features of the primary root are almost totally obliterated. Oil-ducts are quite numerous in the mature root; they are located in the same radii as the mestome bundles and occur in four or five concentric bands. The innermost oil-ducts are to be seen in the leptome itself, the others some distance apart, the outermost being very near the periphery, though not in contact with the cork. It appears as if the ducts of the outermost two bands are mostly pentagonal in transverse sections; while those of the inner are rhombic and somewhat narrower in circumference."

There are thirty-eight oil-ducts in each mericarp in each fruit, twelve on the commisural side, one outside each of the five mestome bundles, and from five to six in the intervals between these. The mericarps are not glabrous, but hairy, consisting of short unicellular pointed hairs which cover the entire dorsal face.

Agri. College, Ames, Ia.

L. H. Pammel.

# CYTOLOGY, EMBRYOLOGY, MICROSCOPICAL METHODS.

AGNES M. CLAYPOLE.

Separates of papers and books on animal biology should be sent for review to Agnes M. Claypole, Sage College, Ithaca, N. Y.

#### CURRENT LITERATURE.

Prenant, A. Cellules Trachéales des Oestres. Archiv. D'Anat. Microscop. 3: 293-336 (Planch. 15-16), 1900. Material used for this investigation was taken, living, from the stomach of the horse, and consisted in larvæ of the

bot-fly (Gastrophilus equi). Examining these larvæ an area of red coloration is always seen at the posterior extremity of the animal. On dissection two bodies are found, one on each side of the digestive tube, which are white and opaque in their anterior three-quarters, and lobulated in structure. In the posterior part they are more granular, and of a reddish color, varying to a purple-red. This part of the organ is provisionally called the "organe rouge." This organ is found to be composed of a number of large oval cells, surrounded thickly by tracheæ, which branch and finally enter into the interior of the cell. Such cells are called "tracheal cells" to distinguish them from the fat cells of the anterior part of the organ. Besides using fresh material, organs were fixed in Flemming's stronger solution; Mann's fluid (picric acid, sat. sol., 10 pts.; sublimate sat. sol., 10 pts.; formol, 5 pts.); formo-picric sol. of Bouin (sat. sol. picric acid, 75 vols.; formol 25; glacial acetic acid, 5 pts.); platinum mixture of Bouin (platinic chloride of 1 per cent. sol., 10 pts.; sat. sol. picric acid, 20 pts.; formol, 10 pts.; or platinic chloride of 1 per cent. sol., 20 pts.; sat. sol of sublimate, 20 pts.; formol, 10 pts.; acetic or formic acid, 2 to 5 pts.); Weigert's neuroglia fluid, consisting of 5 per cent. sol, of acetate of copper, 5 per cent. acetic acid, chrome alum 5 per cent., and 10 per cent. formol; saline saturated sublimate solution; Golgi's fluid.

Of these the formo-picric of Weigert, and Flemming's, gave the best results. The stains used were Benda's safranin and light green, Flemming's triple safraningentian-orange, Mann's blue of toluidin-eosin; especially good results were given by Heidenhain's iron hæmatoxylin after the picric-formol fixative. All sections were cut in paraffin. The author summarizes his results as follows: As before stated, there is present in the larvæ of Gastrophilus equi, Fabr. or pecorum, Fabr. but not in those of Hypoderma bovis L. and Cephalomyia ovis L., an organ occupying the posterior fourth of the animal, having a characteristic red color. This organ has anatomical continuity with the fat body. It is composed of large cells, between which the tracheæ branch and subdivide. The smallest branches penetrate to the interior of these "tracheal cells." Nothing further could be observed as to the ends of the branches beyond fine subdivision. The cytoplasm of the cells is distinguished from the tracheal branches by the different form and stain affinity of its filaments, which come into close relation with the walls of the tracheæ, but do not represent the fine continuations of these tubes. These tracheal cells pass gradually over into adipose cells in the transition region of the organ. This transition is effected by filling the tracheal cell with fat globules and the reduction of the intracellular trachea. Independent of this tracheal organ, certain irregular subcutaneous tracheal cells are found in certain regions of the body. The reason for this specialization is considered to lie in the peculiar habitat of the larvæ, since closely related forms living under different conditions show no such structures. It is an example of limited adaptation. Physiologically, the function of these cells is respiratory, and hence the cells are really "enocytes," differing however from the latter in a red instead of a yellow coloration. The transformation of these tracheal cells into fat cells argues for their "enocytic" nature, and they represent the first step in respiratory differentiation. They are abundantly supplied with oxygen, and in consequence easily elaborate fatty granules; hence the functions of the two parts of this organ are not distinct, but successive.

Zollikofer, R. Kammerfärbung der Leucocyten. Zeit. f. wiss. Mikros. u. f. Mik. Techn.17: 313-321, 1900.

In the study of leucocytes two objects are in view, the fixation of the whole mass of them and the differentiation of

this mass into its different kinds. In order that the study may be done in a counting chamber, it is necessary to mix the blood and the staining fluid, and to have this mixture take place in one pipette. To determine the numerical relations of the different kinds of leucocytes on a cover-glass preparation is impossible, since there is an unequal distribution of the kinds. Lymphocytes are found in thick places of the film, and are rare or crushed in thin places. No such objection can be made to a film of blood in a counting cell. The thing needed was a diluting fluid for this "staining-chamber" which would render the red corpuscles invisible and stain the white differentially. Thin aqueous formalin solution answers the first requirement, and for a stain a mixture of eosin and methylin blue (eosin W. G. and methylin blue B. x. of Grübler, Leipzig) was most satisfactory in the following composition: Eosin W. G. 0.05, concentrated formalin 1.0, distilled water 100.00; methylin blue 0.05, concentrated formalin 1.0, distilled water 100.00. These solutions must be filtered; the

formalin mixture needs to be kept in the dark, and a dark glass dropper was used. About equal parts of the two liquids were taken. A Thoma-Zeiss pipette was filled with blood to 0.5 mark, and filled to 1.20 with the mixture; 1.10 does not completely destroy the red cells. After five minutes, the chamber is filled from the pipette, and the white cells are allowed to settle. Then the blood plates are found to be arranged in characteristic masses, and stain a light gray-blue; the erythrocytes are destroyed; nucleated red cells are sometimes recognizable by their greenish "discoplasm." Malaria plasmodia stain blue, but their recognition is uncertain. Leucocytes are stained as to both nuclei and granules. Eosinophil granules are clearly outlined, and the nuclei remain bright. Neutrophil granules are gray-violet. Most cell granules are unstained. The mononuclear or ungranulated, leucocytes of normal blood are homogenous, with faintly blue cytoplasm and varying nuclei. Nuclei of the larger lymphocytes are clearer, and light violet, the others bright blue and oval. The nuclei of granulated leucocytes stain lightly. The granulated mononuclear, or "Mark" cells, are conspicuous by their size and varying form of nuclei. These are principally recognized by their granules and the size of the nucleus. For a counting chamber, Elzholz's (Reichert) was used. This has a capacity of 0.9 cubic millimeter. The blood is diluted twenty times, and the whole field is counted. The contained number is multiplied by  $\frac{200}{9}$  or 22.222. Many cover-glass stained preparations were also studied. Preparations were stained for a few minutes in eosin, and for half a minute in methylen blue diluted five times with water. Careful fixation is necessary for good results in staining; heating in a hot chamber at 115° for an hour, or for a few minutes at 120-125°, gives good results for both red and white cells. The triacid stain was used when it was desired to stain the neutrophil granules. The mixtures given above afford excellent results not only on blood but also on sputum, pus, and other secretions.

Lewinson, J. Zur Methode der Fettfärbung Zeitschr. f. wiss. Mikros. u. f. Mikr. Tech. 17: 321-326, 1900.

Osmic acid is the usual fixation and staining fluid for fats, but it has several disadvantages. It is expensive, it fixes

but a small part of the tissue put into it or any of the liquids in which it is an active agent. Any fat near the middle of the tissue remains unfixed and unstained. The stain of osmic acid is very often of short duration, and it is almost impossible to use other stains after this fixative. Experimenting on myelinic fibers, a hæmatoxylin method of Wolters' was used. By modifications it was found that other tissues than myelinic fibers took this stain. tried concentrated nuclear stain, warmed, after definite fixation methods, to see if any result in staining fat could be obtained. A concentrated solution of methylen blue in 2 to 5 per cent. salt solution, and hæmatoxylin in acetic acid, were first tried on objects fixed in different fluids. Celloidin sections of the ovary of a rabbit fixed in picric acid were stained in such a warmed solution of methylen blue as described above for 10 to 15 minutes. After decolorizing with weak aqueous hydrochloric acid and counterstaining with alcoholic picric acid, the following results: Nuclei of the cells are blue, protoplasm yellow-green, and the connective tissue is violet. The fat in the follicles takes the form of small, dark, almost black fat-corpuscles. For a modification of Wolters' method the

tissues are fixed in Müller's fluid; the object can have a large surface, but should not be thick. From the fixation fluid the tissue is put into 70 per cent, alcohol, as treatment with weak alcohol and water renders the fat unstainable. The celloidin sections are put in the stain for twelve hours at a temperature of 40°C. A 2 per cent. solution of Kultschitzki's hæmatoxylin (hæmatoxylin 2 gms., dissolved in a little absolute alcohol added to 100 c. c. of 2 per cent. acetic acid). When the mixture, which is at first yellow, becomes red, it is ready for use. The principal point is good decolorization, only well bleached preparations show the fat clearly. The whole process is as follows: (1) Fix in Müller's fluid 2 to 6 weeks, depending on the size of the object; wash out in 70 to 85 per cent. alcohol, etc., imbed in celloidin. (2) Cut sections 10 to 15 mikrons thick, and put them directly from alcohol into the stain for twelve hours at a temperature of 40°C. (3) Wash out with water. (4) Wash in a 1 per cent. solution of potassium permanganate 10 to 15 minutes. (5) Wash in water. (6) Treat with a 2 per cent. solution of oxalic acid, or a mixture of two parts of 2 per cent. oxalic acid to one of 2 per cent. solution of potassium sulphate, for five minutes. Should the preparation show a yellow or gray-black color, return it to the potassium permanganate, then pass to the oxalic acid. If no fat is present the sections lose their color entirely; if fat is there the sections are light ash-gray to an intense gray-violet, depending on the amount present. In this way fat is shown on a colorless background in gray-violet fat globules. If it is desired to stain the nuclei and protoplasm of the cells, a counterstain of concentrated carmin solution may be used as follows: (1) The sections decolorized in oxalic acid are washed in water and left in an ammoniacal solution of borax carmin for twentyfour hours. (2) Treated in acid alcohol (1 per cent. in 70 per cent. alcohol) for two minutes. (3) Sat. alcoholic sol. of picric acid for one minute. (4) 85 per cent. alcohol, absolute, xylol or origanum oil, balsam. The fat is now dark blue, almost black; nuclei, red; protoplasm, yellow. This method is valuable for four reasons: (1) Fat is clearly differentiated to the smallest particle. (2) This fatstain is very lasting; preparations remain good for several months. fluid is an easily available fixation fluid. (4) The method is both inexpensive and simple, requiring no complicated technique. A. M. C.

#### RECENT LITERATURE.

Lavdowsky, M. Ueber eine Chromsublimatverbindung und ihre histologische Anwendung, unter anderem auch zur Restauration älterer Objecte. Zeit. f. wiss. Mikros. u. f. Mikros. Technik. 17: 301-311, 1900.

Weber, A. Contribution à l'étude de la métamérie du cerveau antérieur chez quelques Oiseaux. Archiv. D'Anat. Micros. 3: 369-424, 2 pls., 1900. Nicolas, A. Recherches sur l'embryologie des Reptiles. Contribution à l'ètude de la Fécondation chez l'Orvet. Archiv. D'Anat. Micros. 3: 456-489, 1 pl., 1900.

Goodrich, E. S. Nephridia of Polychæta. Quart. Jour. Micr. Sci. 43: 609-748, 6 pls., 1000.

Yasuda, A. Adaptution of Infusorians to Concentrated Solutions. Jour. Coll. Sci. Tokyo. 13: 101-140, 3 pls., 1900.

#### NORMAL AND PATHOLOGICAL HISTOLOGY.

JOSEPH H. PRATT.

Harvard University Medical School, Boston, Mass.; to whom all books and papers on these subjects should be sent for review.

Bielschowsky and Plien. Zur Technik der Nervenzellenfärbung. Neurologisches Centralblatt, 19: 1441, 1900. Ehrlich and Lazarus introduced the use of cresyl-violet for staining the basophilic granules of mast-cells. Lit-

ten has used the same anilin dye for coloring the basophilic granules which are found in the red blood corpuscles in cases of anæmia. The writers of this article have found cresyl-violet a good stain for the chromophilic substance of the nervecells. They used chiefly the preparation which bears the trade name "cresyl-violet R. R."

In composition this new stain is probably related to methylen blue; in staining properties it resembles thionin or toluidin blue, but is superior to them, chiefly because the preparations are more permanent, and as dilute solutions are employed it is more economical. Another advantage, which the writers claim, is that the sections are never lost in the dilute transparent solution of cresyl-violet.

The best results were obtained with the following method:

- 1. Harden in alcohol or formalin.
- 2. Imbed in celloidin.
- 3. Stain in a thin aqueous solution of cresyl-violet for 24 hours. It is sufficient to add six to ten drops of a concentrated aqueous solution to 50 c.c. of water.
  - 4. Wash quickly in water.
- 5. Dehydrate in a series of alcohols of increasing strength. The alcohol, by removing excess of color from the diffusely stained sections, differentiates the gray and white matter of the central nervous system.
  - 6. Clear in oil of cajeput.
  - 7. Xylol.
  - 8. Mount in Canada balsam.

Equally good results are obtained after imbedding in paraffin. When a quick method is desirable, a concentrated solution of the stain may be used.

Cresyl-violet gives a metachromatic effect with amyloid, coloring amyloid substance bright blue, the remainder of the section violet.

J. H. P.

Krompecher. Glandlike Carcinoma of Epidermic Origin. Ziegler's Beiträge, 28: 1, 1900.

Krompecher describes a peculiar type of tumor of the skin, to which he gives the name "carcinoma epithelialeade-

noides." He believes that the gross and histological appearances are sufficiently characteristic to establish it as a distinct group.

Braun, in 1892, studied this class of tumors, and regarded them as endotheliomata. Krompecher asserts that the diagnosis of endothelioma can be made only when the origin of the tumor-masses from the endothelium of the larger

lymph-spaces can be directly traced, or when the tumor is found in places, such as bones and lymph-nodes, where epithelium is lacking, and the structure of the tumor corresponds to that of undoubted endotheliomata. Braun did not fulfill these requirements of Krompecher. He based his diagnosis on the difference of structure as compared with ordinary epidermoid carcinomata; on the absence of epithelial pearls; and especially on the lack of any connection between the tumor and the skin.

Krompecher studied thirty-three cases. The tumors occurred on various parts of the body. By means of serial sections, he demonstrated the connection of these tumors with the surface epithelium, thus proving their epithelial origin. The striking feature of these tumors is their microscopic structure. While the epidermoid cancer is composed of the cylindrical cells of the stratum Malpighii, and of polygonal prickle cells, which by cornification give rise to epithelial pearls, the group of tumors under consideration is distinguished by the fact that only the cylindrical layer of the stratum Malpighii proliferates. The cells retain their embryonic character. The tumor consists of nests of high cylindrical cells, which stain intensely. There is no formation of epithelial pearls.

J. H. P.

Wright, J. H. A Case of Multiple Myeloma.
Trans. Assoc. Am. Phys. 15: 137, 1900.
Wright defines multiple myeloma as a primary neoplasm of the bone marrow,

affecting chiefly the sternum, the ribs, the vertebræ, and the skull; the substance of the bone being more or less extensively replaced by the tumor tissue. The affection was first recognized by von Rustizky in 1873. It is a rare condition. Less than twenty cases have been reported. The association of albumosuria with multiple myeloma is an interesting feature, and an aid in diagnosis. In the case studied by the writer, the tissues were hardened in Zenker's fluid and in Flemming's solution. The sections were stained in various ways, but eosin and Unna's alkaline methylen blue solution, and fuchsin, either alone or in combination with aurantia, were found most satisfactory.

The tumor is chiefly made up of small cells closely crowded together. Most of the cells have all the appearances of plasma cells, except that the cytoplasm does not in all cases show a marked affinity for methylen blue, as does the typical plasma cell. Wright holds that these cells are plasma cells, and their deviations from the type of the parent cell are quite analagous to those seen in the cells of other neoplasms.

The author found that plasma cells are a normal constituent of the red marrow, and he concludes that the tumor arose from an abnormal proliferation of these cells. Hence his case of multiple myeloma is to be regarded as a neoplasm originating not in the red marrow cells collectively, but in only one of the varieties of the cells of the red marrow, namely, the plasma cells.

J. H. P.

#### GENERAL PHYSIOLOGY.

RAYMOND PEARL.

Books and papers for review should be sent to Raymond Pearl, Zoölogical Laboratory, University of Michigan, Ann Arbor, Mich.

Gamble, F. W., and Keeble, F. W. Hippolyte varians: a Study in Colour Change. Q. J. Mic. Sci. N. S. 43: 589-698, pl. 32-36, 1900.

This paper deals with the color changes shown by the prawn Hippolyte varians, which lives in shallow water clinging to

seaweeds and zoöphytes, in relation to different environmental conditions and to stimulation by light. It has been often noted that Hippolyte shows a very remarkable similarity in its coloration to its surroundings, and it was the purpose of the authors to determine under exact experimental conditions what the nature of these adaptive color changes was, and how they were brought about. As an introduction a description is given of the different natural varieties of this prawn and of the condition of the chromatophores associated with these varieties. Uniform brown, pink, red, and green adult specimens were collected, while among immature individuals "red liners," "black liners," "green liners" and yellow barred specimens were common. The "liners" are animals which are transversely striped with the color indicated. The pigments are contained in chromatophores which lie under the epidermis in the connective tissue and muscles and about the alimentary canal and blood vessels. The chromatophore itself consists of a central body from which diverge a number of fine, ramifying, hollow tubes. In these tubes and the central body are contained the pigments which give the color to the animal. There are three pigments, red, yellow and blue, and color changes are caused by the movement of these pigments in the There is a layer of chromatophores in the connective tissue just beneath the epidermis, the processes of which form a close meshwork about the clear transparent cells of the epidermis itself. To the combination of the three pigments in this epidermal meshwork is due the color of the animal as a whole. For example, in a green prawn the tubes of the meshwork are found to contain both yellow and blue pigment side by side.

The animal exhibits three sorts of color changes: (a) slow, sympathetic changes of color accompanying changes in the color of the weed to which the individual is attached; (b) rapid color changes caused by changes in light intensity; (c) periodic nocturnal color changes. (a) In regard to adaptive color changes in response to changes of the weeds, it was found that the animals were capable of only very slow sympathetic changes. The adaptations observed in nature are the result of the selection by the animal of those weeds whose color most closely matches their own. (b) The intensity of illumination has a pronounced effect on the color of the animals and this effect is produced in a very short time. In high light, or in low light scattered evenly from the surface of the containing dish, there is a retraction of the red pigment and an evolution of the blue and the yellow, producing a green coloration of the animal; while in low light absorbed by the walls of the vessel the red remains expanded. The color quality of the light has no effect on the color of the animal provided the

intensity remains the same. (c.) It was found that at evening the prawns change regularly and uniformly from their diurnal color to a deep, transparent blue. This blue color passes away in the morning and the diurnal color of the previous day reappears. This nocturnal change with its associated diurnal recovery is a periodic phenomenon. The nocturnal blue appears at certain intervals even if the illumination is kept constant for several days at a time, and on the other hand the recovery of the diurnal color occurs regularly in specimens kept in the dark during similar long periods. Blinded prawns exhibit the same periodicity. The general physiological condition of the animals is very different during the night and the day.

Other experiments showed that the condition of the chromatophores at any time is the result of impulses passing to them from the central nervous system. Color changes can be induced by a variety of stimuli such as temperature, chemicals, electricity, etc., which affect the nervous system. Removal of the eyes causes a change in the impulses going to the central nervous system and hence a change in the color. The nocturnal condition is a result of a periodicity in the action of the nervous system. The relation of the nocturnal color of these prawns to the color of deep sea animals is discussed.

This paper is an important contribution to the physiology of coloration. Unfortunately in the space of a brief review it has been impossible to do more than mention some of the most significant points in the great amount of interesting detail presented in the work. The admirably executed plates are a feature of the paper. The experimental methods described are numerous and valuable.

R. P.

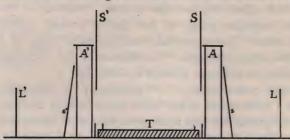
Yerkes, R. M. Reactions of Entomostraca to Stimulation by Light. II. Reactions of Daphnia and Cypris. Amer. Jour. Physiol. 4: 405-422, 1900.

The purposes of the investigation were to determine the relation between the rate of movement and the intensity of

light; to determine whether there is a reversal of the phototactic reaction of Daphnia and Cypris under certain conditions; and finally to study the effect of other stimuli on the light reactions. In regard to the first point it was found that Daphnia moved only slightly faster as the intensity of the light increased, . while Cypris showed a still less definite relation between rate and intensity than Daphnia. No very conclusive results came from the experiments devised to test the question of reversal of phototaxis from positive to negative and vice versa. Daphnia usually gives a positive reponse which could be maintained indefinitely by changing the direction of the light. In some cases it was possible to change this positive reaction into a weak negative one by taking the animals up in a pipette as described by Towle (Amer. Jour. Physiol. 3: 345-365), but this result was not constant. The reverse change from negative to positive was not obtained, owing apparently to lack of negatively phototactic animals on which to experiment. In the case of Cypris, negative animals were made positive by contact with the sides of the pipette. Raising the temperature does not affect the phototaxis of these crustaceans. Sudden illumination of the animals from above in such a way that the directive influence of the light is excluded does not cause any change in the direction of swimming. In one set of experiments some strong HCl was put into the trough at the end nearest the source of light. The

animals swam towards the light until they encountered the acid solution, and then instead of turning back stayed there till they were killed. In conclusion the author gives a rather unconvincing answer to certain criticisms of his earlier work.

In the paper several useful pieces of apparatus for phototaxis work are described. The method of changing the direction of the light rays impinging on the animal without disturbing any of the other conditions seems especially valuable and will be described in detail. "A tin trough  $8 \times \frac{1}{2} \times \frac{1}{4}$  inches (T) mounted on a wooden base was painted dead black; at either end of this trough a glass box, A, A', containing alum solution was placed. Screens, S, S', were arranged so that side rays and reflected light were cut off, and the trough was illuminated exclusively by rays parallel with its long axis coming through holes six inches high and two inches wide cut in the screens, S, S'. At either



end, ten inches from S and S' respectively, was a Welsbach burner, L, L'. For observations this apparatus was set up in a dark room. After the trough had been filled with water and the screens s, s', which shut off all light, had been

placed in position, an animal was carefully dropped into the middle of T. One of the screens (s) was then removed and the animal responded usually with a + reaction,—it moved toward the end from which the screen had been removed, that is, toward the light. As soon as the animal came within two centimeters of the + end of the trough, s was quickly replaced and s' removed, thus giving light from the opposite direction without the inconvenience of moving the burner. By this means it could easily be observed whether the response was continued as before or reversed."

Putter, A. Studien über Thigmotaxis bei Protisten. Arch. Anat. u. Physiol. Physiol. Abth. Suppl. Bd. 1900: pp. 243-302. The author deals in a thorough and exhaustive way with the effect of contact with solid bodies on the reactions

of the Protozoa. After a brief historical introduction and description of methods employed, the reactions of a large number of Protozoa, including nearly all the main groups from the rhizopods to the hypotrichous ciliates, are described in detail. Positive and negative forms of thigmotaxis are distinguished according as the animal remains in contact with a solid body which it encounters, or moves away from it. The positive reaction displays two forms or factors. The first factor is the one which affects the locomotor organs (pseudopodia, flagella or cilia) and results in a lessening or inhibition of their movement. The second factor in the thigmotaxis is the secretion of a sticky slime which helps to fasten the animal to solid bodies. This secretion factor is very evident among the rhizopods, less apparent among the flagellates and ciliates where the first factor is most important, and finally it is the most essential phenomenon in the thigmo-

taxis of Oscillaria, diatoms and desmids, and the Gregarinidæ. It is evident that these phenomena of thigmotaxis are very important in the life of the Protozoa.

This importance is well shown by the effect of the thigmotaxis on the reactions to other stimuli. The other reactions studied were the electrotactic and the thermotactic. In regard to the electrotaxis it was found that among some of the ciliates, individuals which were kathodically electrotactic when swimming freely through the water, when in contact with a solid body (i. e., thigmotactic) oriented themselves more or less transversely to the direction of the current with the oral side of the body towards the kathode. This transverse orientation was investigated in a number of forms. It is evidently the same reaction as that which has been described by the reviewer (Amer. Jour. Physiol. 3: 96-123) and explained as due to the conflict between two sets of ciliary activities. It is now shown to be also in part the result of the thigmotaxis of the animal. The author confirms previous investigators as to the reversal of the cilia on the kathode side of the body during the action of the current. The permanent transverse electrotaxis of Spirostomum is thought to be a result of the thigmotaxis of the posterior end of the body. Professor Loeb's theory of the action of the external electrolytes in electrotaxis is thoroughly examined and strong evidence against it is presented. The effect of heat or cold is different according as the animal is, or is not, in contact with a solid body. Many forms (Euglena, Chilodon, Stylonychia, Spirostomum and others) cannot be made to leave the bottom by heating. They die while still thigmotactic.

The reactions of Stylonychia mytilus are described in more detail than those of any other form and some interesting curves are given showing the relative activities of the different groups of cilia at different temperatures. All the cilia show maximal activity at two widely separated temperatures (5–10° and 25–35°C.) while the minimal activity of all is between  $15^{\circ}$  and  $20^{\circ}$ C.

This excellent piece of work puts our knowledge of another of the reactions of the Protozoa on a firm basis.

R. P.

Delage, Y., and Delage, M. Sur les relations entre le constitution chimique des produits sexuels et celle des solutions capables de dèterminer la parthénogenèse. C. R. Ac. Sci. Paris, 131: 1227-1220, 1900.

In this note are presented the results of some chemical analyses of the sexual products of the male and female in the sea urchin, Strongylocentrotus lividus. The

starting point of the work is the idea that if, as has been stated, it is the Mg-ion which causes the artificial parthenogenetic development of the egg, and if normal development is the result of the same sort of a process, analysis ought to show a greater proportionate amount of this salt in the sperms than in the eggs. This was not found to be the case. The magnesium content of the products of both sexes is essentially the same, so that normal fertilization cannot depend merely on the bringing of more of this salt into the egg by the sperm. This result in no way affects Professor Læb's later views, which point to osmotic pressure as the essential factor in the production of artificial parthenogenesis.

#### CURRENT BACTERIOLOGICAL LITERATURE.

H. W. CONN.

Separates of papers and books on bacteriology should be sent for review to H. W. Conn, Wesleyan University, Middletown, Conn.

Jensen. Studien über die Enzyme im Kase. Cent. f. Bac. II. 6: 734, 1900.

Babcock and Russell. Relation of the Enzymes of Rennet to Ripening of Cheddar Cheese. Cent. f. Bac. II. 6: 817, 1900. (See also seventeenth annual report of Agri. Expt. Sta. of Wis.)

Babcock and Russell. Causes Operative in the Formation of Silage. Seventeenth An. Rep. of Agri. Expt. Sta. of Wis., 1900.

Behrens. Ueber die oxydierenden Bestandtheile und die Fermentation des Deutschen Tabaks. Cent. f. Bac. II. 7: 1, 1901. The question whether various fermentative processes in nature are to be ascribed properly to the action of micro-organisms or to the action of enzymes, has, in late years, become a somewhat burning one with our bacteriologists and chemists. In large degree the question reduces itself simply to determining whether the enzymes, which are the direct cause of the action,

are produced by bacteria or from some other source. The four papers here referred to discuss different aspects of this problem. It has been shown by Babcock and Russell that fresh milk contains an enzyme which they have named galactase. They believe that this enzyme, rather than micro-organisms, plays the important part in the ripening of cheese. The first of the articles here referred to contains an especially careful series of experiments to test this conclusion. As the result of a long series of most careful experiments, Jensen concludes, in brief, that in the ripening of soft cheeses the effect is produced, (1) by enzymes which are produced by yeasts and bacteria growing on the surface of the cheese; and (2) by enzymes in the center of the cheese which are not derived from bacteria growth, but rather from the rennet which was added to curdle the casein, the enzyme in this case being pepsin. The ripening of hard cheeses depends partly upon the action of an enzyme produced throughout the mass as the result of bacteria, and partly, especially in the early part of the ripening, upon the galactase, which, as Babcock and Russell have shown, is present in the fresh milk.

In the second article Babcock and Russell test, by an entirely different line of experiments, the question whether the pepsin present in the rennet has an important agency in the ripening of cheese. The conclusion they reached is essentially identical with that of Jensen, namely, that the ripening of cheese is dependent in considerable degree upon the pepsin present in the rennet. The agency of bacteria in the ripening of cheeses is not especially studied by these authors.

The third paper records a series of experiments to determine whether the production of silage is, as has previously been believed, the result of the growth of micro-organisms. The authors reached the conclusion that micro-organisms have nothing whatsoever to do with the production of normal silage. Both the initial heating and the subsequent ripening of silage are due to entirely different agents. The production of silage, the authors believe, is due, (1) to the

respiratory processes of plant tissues which continue for some time in the silage after the silo is packed, thus producing the initial heating; (2) to the presence of enzymes which are liberated from the plant cells after the death of the plant tissue.

Micro-organisms, the authors believe, only injure the silage and are of no significance in a properly constructed silo.

The paper by Behrens deals with the question of the fermentation of tobacco, which has been regarded as due to micro-organisms, but which Loew has somewhat recently insisted is the result of enzymes formed in the tobacco leaves. Behrens has tested Loew's conclusion and was able to isolate from the leaves of German tobacco the same chemical products referred to by Loew. After making a somewhat careful study of them and their action, he reaches, however, quite different conclusions from those of Loew. His conclusions are, briefly, that these bodies (oxydase, peroxydase) are formed in tobacco leaves. He is doubtful as to whether they are properly to be called enzymes, and is convinced from his experiments that they cannot be the cause of the tobacco fermentation, inasmuch as they disappear from the leaves before the important fermentation takes place. His experiments further show that these oxydases will not produce ammonia from nicotine, a phenomenon of tobacco fermentation which he attributes to bacteria. He finds, also, that micro-organisms will grow in tobacco when the amount of water is not over 25 per cent., contrary to Loew's claims, and is, therefore, convinced that the chief factor in the proper tobacco fermentation is due to bacteria growth rather than to these chemical bodies produced in the tobacco leaves. H. W. C.

Harrison. Die Lebensdauer des Tuberkel-Bacillus im Käse. Landw. Jahrb. der Schweiz., 1900. Harrison has experimented upon the length of time in which tubercle bacilli remain alive in cheese. His method of

experiment has been to inoculate milk with a considerable quantity of tubercle culture and then to make the milk into cheese in the ordinary fashion. At varying intervals the cheese was tested by inoculation into guinea pigs. These animals were studied both clinically and microscopically. He found that, in Emmenthaler cheese, the tubercle bacilli were dead at the end of 33–40 days, while in Cheddar cheese they might remain alive for 104 days. The conclusion is that neither of these cheeses is a source of danger to man, since they are seldom eaten until they are four months old, or even older.

H. W. C.

Lameris and Harrevelt. Bakterienbefund in Kuhnmilch nach algeheilter Mastitis. Zeit. f. Fl. u. Milch Hyg. 11: 114, 1901. The authors made a study of the bacteria content of some cows' milk which had produced cases of diarrhoea.

Inspection of the source of the milk showed that some of the cows had formerly suffered from mastitis, but had apparently recovered. In the milk, however, there was present a species of streptococcus which is uniformly found and which is really the cause of the intestinal disturbance produced by the use of the milk. Inasmuch, however, as the milk produced these disturbances, even after boiling, and the streptococci were shown to be killed by this temperature, the authors conclude that the trouble arose from the toxines developed in the milk by the streptococcus rather than by the direct action of the organisms.

H. W. C.

#### Medical Notes.

THE EPILEPSY PARASITE.—The short paper published in the December number of the JOURNAL does not represent the present status of this parasite, and in justice to myself and the cause of science a little more should be said about it. That a parasite, represented in the cut accompanying the December article, is the cause of some forms of reflex epilepsy, is an undisputed fact. In the first case at Chester, Illinois, the boy was cured by the permanent removal of the parasite, and has remained cured. Three other cases are known to the writer where the parasite was found. Only one of these three was known to the writer in detail, and the removal of the parasites cured this case as in the first.

The parasite was found to be new to science, and a description of it was published in the September number, 1900, of the Canadian Entomologist, under the name of Gastrophilus epilepsalis. In the September number of the Alkaloidal Clinic of Chicago was published a more extended account of what was known of the three cases that had been found then. This article treated the subject more from a pathological standpoint than the article in the Canadian Entomologist. Since that, one other case has been found in Kentucky, where the parasite was connected with epilepsy.

The parasite, instead of being a Nematoid worm, is the larva of a fly, related to the horse bot-fly, *Gastrophilus equi*. The adult fly has not yet been recognized, nor has it been ascertained definitely how it first gains entrance to the system. In the investigation of this parasite, two other fly parasites infesting the human intestines have been found by the writer that the books do not tell us about, one a species of *Eristalis*, of the family Syrphidæ, and the other a species of *Sarcophaga*, of the family Sarcophagidæ.

I should like to get specimens of intestinal parasites, and have correspondence relative to the effect of such parasites on the system of the host.

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Dr. Klett of Würtemberg has recently made important researches upon the phenomena of anaërobic life. In his investigations on the production of sporeless anthrax outside the living body, Dr. Klett found that if nitrogen is substituted for air in the anaërobic conditions, the growth of the organism is not impaired, and spores develop as freely as under ordinary conditions. If, however, hydrogen is substituted in place of air, no spores develop providing the medium is such as to permit intimate contact of gas with the culture. These results would indicate that absence of oxygen is responsible for non-production of spores in anaërobic cultivation of anthrax.

At a meeting of Pathologists and Bacteriologists in New York on January 26th, an American association was organized. The officers elected were: Dr. W. T. Councilman, president; Dr. H. C. Ernst, secretary; Dr. Eugene Hodenpyl, treasurer. The first regular meeting will be held in Boston on April 5th.



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I left off coffee that day at lunch and had a cup of Postum. It was made good and had a rich, dark color, with a delicious flavor that I could not tell from regular coffee. It pleased the eye, smell, and palate, so I had it each day at the restaurant for the noonday lunch, and discovered a decided improvement in my condition, but it was not until I left off coffee for breakfast and used Postum in its place that real relief set in. Now I am free from gastritis, headaches, and fully appreciate the value of the 'nerve ease.' No more trembling hands and no more nervous prostration. I am well, and feel that I should say to others who are being poisoned by a beverage that they do not suspect, 'coffee,' 'Make the change before the poison works destruction in you.'''

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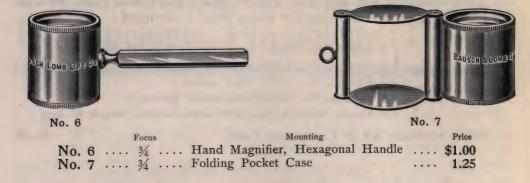
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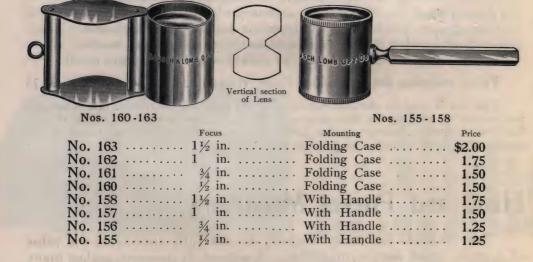
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These magnifiers, often called linen testers, fold up to occupy the smallest possible space. The lens gives about ten diameters magnification, and is focused on the opening in the base so that the magnifier is

simply set over the object. The several sizes of openings are useful in examinations of linen and other fabrics.

	Size of Opening, Mm.	Shape of Opening	Price
No. 141	25 x 25	Square	\$2.00
No. 141½	12 x 12	Square	
No. 142	6 x 12	Rectangular	
No. 143	6 x 6	Square	
No. 143½	6	Circular	

### Folding Pocket Lenses

Our pocket magnifiers have always been considered the standard. They have superseded those of foreign manufacture mounted in horn, etc., as the vulcanite which we employ for mountings is much preferred on account of its greater permanence, lightness, and neat appearance. The lenses used are accurately ground and give good results.

#### VULCANITE MOUNTING

OVAL FORM



No. 50 Magnifier



No. 51 Magnifier
Actual size

		Diam.	of Lenses in	Mm.	Price
No.	50		18		\$ .20
No.	51		15, 18		.40
No.	56		25		.30
No.	57		21, 25		.50
No.	62		30		.40
No.	63		28, 30		.65
No.	68		37		.50
No.	69		30, 37		.80
No.	74		43		.60
No.	75		37, 43		1.00
No.	78		50		.75
No	70		43 50		1 25

#### BELLOWS SHAPED

	Dia	m. of Lenses in	Mm.	Price
No. 101		18		\$ .20
No. 102		15, 18		.35
No. 103		12, 15, 18		.50
No. 110		21		.25
No. 111		18, 21		.40
No. 112		15, 18, 21		.60
No. 119		25		.30
No. 120		21, 25	•, • •	.50
No. 121		18, 21, 25		.80

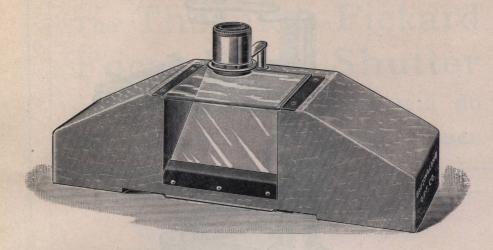


No. 101 Magnifier
Actual size



No. 103 Magnifier

### Barnes Dissecting Microscope

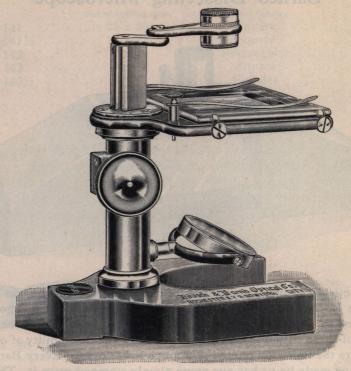


This practical and inexpensive form of dissecting microscope was designed by Prof. Charles R. Barnes, Botanical Department, University of Chicago. Its popularity has been so great that many copies of it have been put on the market. A comparison of these with our instrument will show the desirability of our construction even were the imitation offered at a much lower price. Every Barnes Dissecting Microscope made by us has our name plainly stamped upon it. The T and TT Microscopes will be found extremely well suited for class use in elementary botany, zoölogy, etc. The body of the stand is of neatly finished light wood, and is shaped to form hand rests. The stage is extra large and easily removed for cleaning. The mirror is as large as the stage, giving effective illumination. A plate, with a black and white side, is stowed beneath the stand, and may be laid over the mirror if a black or white background is desired. The lens carrier slides in a metal sleeve, which is firmly fixed in the wooden base, permitting focusing with sufficient accuracy for powers required. Doublet lenses, having much larger, flatter field, and better definition than the simple lenses usually furnished, are listed with this microscope and form a most desirable equipment at the price. Coddington or Aplanatic Triplet lenses may be used with equal facility, and are more desirable where their cost is not prohibitive.

In many laboratories where elementary work is done there is no convenient receptacle for the scalpel, tweezers, dissecting needles, etc. To provide for this we have added to the T Microscope an iron base hinged to the wooden stand so that it forms a tray for material and at the same time adds to the stability of the instrument. We would recommend the purchase of this stand, which we designate as the TT, in all cases where cost will permit.

CALL TO STATE OF THE PARTY OF T	Doublet		Price
No. T1	1 i	n	\$2.50
No. T2	2 in.,	1 in	3.25
No. TT1	1 is	n	3.00
No. TT2	2 in.,	1 in	3.75

### Dissecting Microscope—W



This microscope is especially adapted for botanical and zoölogical work, and its extremely moderate price recommends it for individual use. It is constructed to secure great steadiness under manipulation, convenience in working, and durability. The base is heavily japanned, all other parts being nickeled to prevent corrosion. The mirror frame holds a concave mirror and a white plane glass reflector. The stage is extra large, and the entire surface of the thick glass stage plate is available for work, as the spring clips are attached to the metal supporting frame. The stage plate is held in place by spring clips so as to be easily removable for cleaning, etc. Size of stage plate, 75 x 100 mm.

The lens arm is jointed so that the lens may be moved over every part of the stage. The focusing arrangement is by accurate diagonal rack and pinion of very long range, giving great working distance between lens and stage. Aplanatic triplet lenses should always be selected when cost is not prohibitive. Lenses of the foci listed are most generally used. Those of any of the regular foci can be

substituted if preferred.

Each W Microscope is furnished in neat wooden carrying case.

Ing needles, etc. To provide for the	enses and allowed Williams and the Price				
No. W1 1 in.	Doublet \$ 9.75				
	Doublet 10.50				
No. W3 1 in.	Coddington 10.50				
No. W4 1½ in., ¾ in.	Coddington 12.00				
No. W5 1 in.	Aplanatic Triplet 12.50				
	Aplanatic Triplet 16.00				
Folding Wooden Hand Rests, per pair					

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